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SIXGILL SHARK (*HEXANCHUS GRISEUS*) CONSERVATION ECOLOGY PROJECT UPDATE

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## INTRODUCTION

The bluntnose sixgill shark *Hexanchus griseus* is large (up to 5m in length), heavy-bodied shark (500 kg) (Castro, 1983). They are considered primitive sharks because of distinct characteristics that differentiate them from modern sharks, such as sixgill slits and dimorphic top and bottom teeth (Ellis, 1983). Sixgill sharks are a demersal species (Castro 1983, Ebert 1986, Ellis 1983, and Rupp 2001) found in tropical and temperate seas around the globe (Castro 1983, Ebert 1986, Ellis 1983, and Rupp 2001). With the exception of Blue Sharks, sixgill sharks are, perhaps, the most wide-ranging of all shark species (Ebert 2003). Although it is typically a deepwater shark occurring on the continental shelf and upper slope, they may occasionally migrate to shallower waters, and juveniles may occur close to shore (Ebert 1986; Ebert 2003). In the Northern California Current, the occurrence of pre-reproductive sixgill sharks in shallow water appears to vary seasonally. Time-lapse video observations in the Strait of Georgia, British Columbia revealed a higher occurrence of sharks in summer relative to other months (Dunbrack and Zielinski 2003).

Sixgill sharks are viviparous and produce litters of 47-108. Males appear to mature at about 3.1m and females at about 4.2m (Ebert 2003; Ebert 2002), although very little is known about their age of maturity. Little is also known about their growth rate, although Ebert (2003) reports that juveniles double in size in their first year of life. Sixgill sharks appear to feed on a wide range of species including sharks, rays, chimaeras, herring, hake mackerel, halibut and whale carrion (Ebert 1994; Ebert 2003).

The existence of sixgill sharks was an "open secret" among Puget Sound divers, recreational and commercial fishermen, scientists and naturalists, but few people in the general public knew that Puget Sound and Seattle's Elliott Bay contained a well-established, but rarely seen shark population. Anecdotal evidence (acquired from personal reports of divers and fishermen) suggests that Puget Sound's sixgills are long-lived and slow-growing, and appear to have established movement corridors and territories that remain relatively fixed over time. Sixgills prey upon Puget Sound's dogfish, skates and other non- or low-value commercial fishes, and like other apex predators are highly vulnerable to changes in their environment (Ebert 1986).

Worldwide concern over the effects of fishing on shark populations was brought to the forefront of public attention in Seattle by publicity surrounding the catching of several bluntnose sixgill sharks from Elliott Bay during the summer of 2000. These sharks were fished from an area where local divers had reported frequent encounters with known individual sharks in shallow waters (Duwamish Head, Elliott Bay). After the fishing pressure, no sharks were sighted in this area for several months. Eventually sharks were re-sighted in the area but not the individuals previously identified. This apparent direct affect of fishing pressure on the local sixgill population caused concern within the local diving and scientific communities.

State regulators responded to these concerns by placing a temporary closure on the taking of these sharks in Puget Sound. The Seattle Aquarium was instrumental in acting as an informational liaison between the public and the Washington Department of Fish and Wildlife (WDFW) to obtain the closure. Lack of information on the abundance of sixgills in Puget Sound/Georgia Basin, their movement patterns, and basic biological parameters, dictated a cautious approach to allowing harvest of sixgill sharks.

The growing interest and concern about sixgill sharks stimulated the Seattle Aquarium to develop a long term conservation research program to address gaps in the body of scientific knowledge on these animals. The major goals of the sixgill research are twofold: (1) to examine patterns of movement, home ranges, local abundance, and population boundaries through genetics, visual tagging and acoustic tagging and (2) to establish the aquarium as an informational clearing-house for diver sightings, updates on sixgill research, and information on sixgill natural history and conservation through the aquarium's interactive web site.

## HISTORY TO DATE

A joint research team to study Puget Sound's sixgill sharks was established in 2000 to gather the basic biological information consisting of representatives from the University of Washington, the Seattle Aquarium, the Point Defiance Zoo and Aquarium and WDFW. This group was coined the "Southern Working Group" after a workshop held at the Vancouver Aquarium Marine Science Center in November 2000 to coordinate past and existing sixgill shark research efforts. Presentations were made by representatives of Canadian and United States (U.S.) governmental agencies, regional non-government organizations (NGOs, e.g., aquariums), U.S. and Canadian universities and members of the sport diving industry. In March 2001 the Seattle Aquarium chaired a panel session at the Puget Sound Research Conference to set the stage for future directions in sixgill shark research. By May 2001 the WDFW put in place a permanent closure on the recreational and commercial take of sixgill sharks. Contact information was published in the state fishing guidelines asking all persons who encounter sixgill sharks to report their sightings and encounters to the Seattle Aquarium or the Point Defiance Zoo and Aquarium.

Active sixgill research at the Seattle Aquarium began in October of 2001 when the aquarium partnered with Hydrus Productions to test a 'proof of concept' event to see if sixgills could be attracted to the aquarium's location at pier 59. The event was a success with the first shark arriving almost immediately after bait was set under the pier. Buoyed with this initial success, early work to determine lighting, video camera placement, and diving procedures occurred during the spring/summer of 2002. By the fall of 2002, the aquarium had constructed the permanent research station on the seafloor beneath pier 59 providing a protected contact area for researchers. Since the fall of 2002, shark research events have occurred every other month. Initial events focused on video documentation of shark behavior and shark/diver interactions and the sharks' response to contact with a blunt spear. The first successful visible marker tagging and biopsy sampling occurred in June 2003 during documentary filming by National Geographic Television. As of September 2003, the aquarium had successfully tagged 39 sharks with visible Floy tags and collected 22 biopsy tissue samples for genetics. In the fall of 2004 Phil Levin with National Marine Fisheries Service (NMFS) joined the Southern Working group and began collaborating with the aquarium on acoustic tagging. In November 16 2005 we placed our first Vemco coded V32 tag attached externally to the dorsal musculature of a sixgill sharks. In March 2005 we tagged two more sharks and tested a Vemco VRAP (radio acoustic positioning system) deployed just off the aquarium's pier to determine shark locations.

## RESULTS

**Population genetics:**

Polymorphic nuclear microsatellite genetic markers are useful for individual identification and for determining genetic variability, population structure, relatedness and geneflow. Microsatellite markers were developed for sixgill sharks at the aquarium following methods described by Olsen et al. (1998). To date we have 12 sixgill specific markers (SG5, SG10, SG11, SG12, SG13, and SG24, SG25, SG26, SG27, SG28, SG29, SG30, SG31 and SG32 Larson, unpublished data) and one marker borrowed from the lemon shark (*Negaprion brevirostris*, LS 15, Feldman et al., 2001) for a suite of 13 markers. Nuclear DNA is extracted using the QIAamp Dneasy protocol (Qiagen corp., CA) from tissue samples collected using our modified stainless steel pneudart biopsy punch, from fisheries sampling and/or from beach cast animals. To date we have microsatellite data for over 200 sixgills from central and southern Puget Sound for our first 7 microsatellite loci (the other loci are currently being analyzed). The average expected heterozygosity ($H_E$), or genetic diversity, over seven of the 13 loci was 0.76. Departures from Hardy Weinberg expectations were not significant for any loci. This initial level of diversity suggest a relatively large breeding population (sexually mature adults) of at least 7916 individuals or $N_E$ (where $N_E = H_E / 4(\mu)(1 - H_E)$, and $\mu = 10^{-4}$ , the mammalian mutation rate for microsatellites; Waples, 1991). This data suggests that the central and southern Puget Sound sixgill population has not suffered from a recent population crash or bottleneck from fishing pressure or ecological changes. The population size estimate is most likely conservative because sixgill phenotypes have changed little in millions of years (Ebert, 1986), suggesting genetic change may also be relatively slow (such as

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microsatellite mutation rate). The true microsatellite mutation rate within sharks has yet to be determined. These initial population genetics statistics illustrate that these markers and conservation genetics methods may be useful in determining sixgill shark population structure, phylogenetic relationships, identifying individuals, and relatedness analyses.

**Tag returns:**
Since visual tagging began in the summer of 2003 we have tagged 38 sharks visually and acoustically tagged 3 individuals during our bi-monthly research events. Fifteen of those sharks have returned at least once to our research site resulting in a tag return rate of 40.54%. We counted a returned tag as one that had been gone from the research station for more than 24 hours. Time at liberty between tags varied (Figure 1) with all 15 returned sharks returning after an average of 87 days at liberty, four sharks returning after an average of 44 days at liberty for the second time, three sharks returning after an average of 136 days at liberty for the third time and one shark returning for the fourth time after being gone for 232 days.

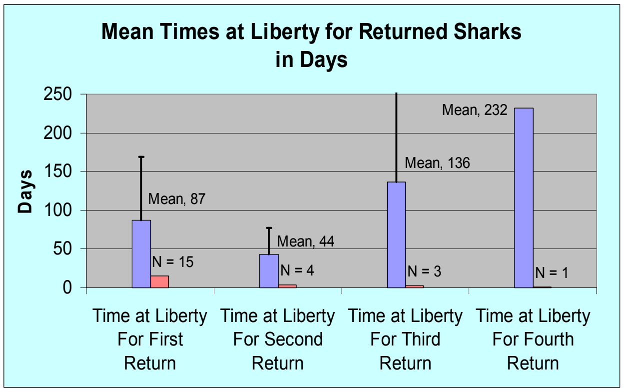


Figure 1. Time at Liberty

Figure 2 summarizes our tagging activity since it began in 2003. The vast majority of animals are tagged and re-sighted during summer months mirroring the anecdotal seasonal abundance of sixgill sightings reported by recreational divers and fishers.

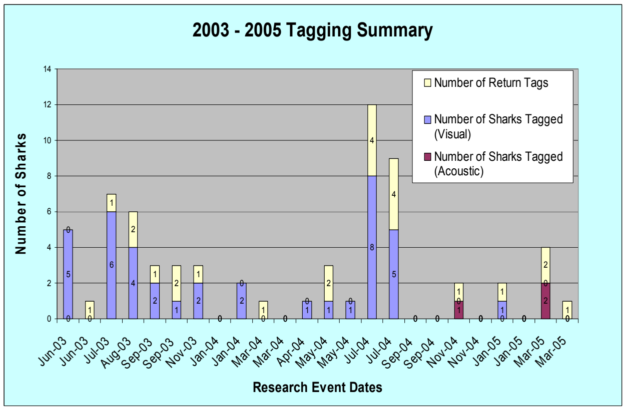


Figure 2. Visual and acoustic tagging summary

## CONCLUSION

The Seattle Aquarium in cooperation with one of Southern Working Group partners, NMFS, will begin active tracking of acoustically tagged sixgill sharks in spring/summer of 2005. We will first begin with boat based tracking of individuals for 48 hours. Once we have sufficient data on individuals and know approximately where their home range is we will move to Chat tags and the Vemco VRAP and chat tag technology to identify shark locations and depth over time without manual active tracking. In addition to visual tagging, acoustic tagging and genetic biopsy sampling research will have to be moved of the end of Pier 59 this summer due to extensive piling replacement construction. Our plan is to continue the bi-monthly research but from either another fixed location or mobile via our research vessel.

The Seattle Aquarium's sixgill shark research program is a long term investigative effort with multiple partners to further the body of knowledge of this exciting apex predator. Studies begun at the aquarium's research site will be expanded geographically and merged with parallel investigations pursued by our partner institutions. The aquarium intends to utilize the sixgill's role as charismatic megafauna to steer public awareness of conservation issues affecting the aquatic ecosystem.

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